

What is claimed is:

1. A method of promoting inhibition of expression of a selected protein comprising:

(a) providing an antisense oligonucleotide targeted  
5 to an RNA encoding a selected protein whose expression is to be inhibited;

(b) allowing said oligonucleotide and said RNA to hybridize to form an oligonucleotide-RNA duplex; and

(c) contacting said oligonucleotide-RNA duplex with  
10 an enriched amount of a mammalian RNase H polypeptide and under conditions in which cleavage of the RNA strand of the oligonucleotide-RNA duplex occurs,

so that inhibition of expression of the selected protein is promoted.

15 2. The method of claim 1 wherein the mammalian RNase H polypeptide is a human RNase H polypaptide.

3. The method of claim 1 wherein the mammalian RNase H polypeptide is an RNase HI polypeptide.

4. The method of claim 1 wherein the mammalian  
20 RNase H polypeptide is an RNase HII polypeptide.

5. The method of claim 1 wherein the mammalian RNase H polypeptide present in enriched amounts is overexpressed or exogenously added.

6. The method of claim 1 wherein the mammalian  
25 RNase H polypeptide present in enriched amounts is an isolated, purified mammalian RNase H polypeptide.

7. The method of claim 1 wherein the mammalian RNase H polypeptide present in enriched amounts is a cloned and expressed mammalian RNase H polypeptide.

8. A method of promoting inhibition of expression  
5 of a selected protein comprising:

(a) providing an antisense oligonucleotide targeted to an RNA encoding a selected protein whose expression is to be inhibited;

(b) allowing said oligonucleotide and said RNA to  
10 hybridize to form an oligonucleotide-RNA duplex; and

(c) contacting said oligonucleotide-RNA duplex with an RNase HI polypeptide having SEQ ID NO: 6, 7, 8, 9 or 11, under conditions in which cleavage of the RNA strand of the oligonucleotide-RNA duplex occurs,

15           whereby inhibition of expression of the selected  
protein is promoted.

9. The method of claim 8 wherein the RNase HI polypeptide is a cloned and expressed RNase HI polypeptide.

10. A method of promoting inhibition of expression  
20 of a selected protein by an antisense oligonucleotide  
targeted to an RNA encoding the selected protein  
comprising:

(a) providing an antisense oligonucleotide targeted to an RNA encoding a selected protein whose expression is to be inhibited;

(b) allowing said oligonucleotide and said RNA to hybridize to form an oligonucleotide-RNA duplex; and

(c) contacting said oligonucleotide-RNA duplex with an RNase HII polypeptide having SEQ ID NO: 1 or 10, under

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conditions in which cleavage of the RNA strand of the oligonucleotide-RNA duplex occurs,

whereby inhibition of expression of the selected protein is promoted.

5           11. The method of claim 10 wherein the RNase HII polypeptide is a cloned and expressed RNase HII polypeptide.

12. A method of eliciting cleavage of a selected cellular RNA target comprising:

10           (a) providing an antisense oligonucleotide targeted to a selected RNA target to be cleaved;

            (b) allowing said oligonucleotide and said RNA target to hybridize to form an oligonucleotide-RNA duplex; and

15           (c) contacting said oligonucleotide-RNA duplex with an enriched amount of a mammalian RNase H polypeptide and under conditions in which cleavage of the RNA strand of the oligonucleotide-RNA duplex occurs,

            so that cleavage of the cellular RNA target is  
20           elicited.

13. The method of claim 12 wherein the mammalian RNase H polypeptide is a human RNase H polypeptide.

14. The method of claim 12 wherein the mammalian RNase H polypeptide is an RNase HI polypeptide.

25           15. The method of claim 12 wherein the mammalian RNase H polypeptide is an RNase HII polypeptide.

16. The method of claim 12 wherein the mammalian RNase H polypeptide present in enriched amounts is overexpressed or exogenously added.

17. The method of claim 12 wherein the mammalian RNase H polypeptide is an isolated, purified mammalian RNase H polypeptide.

18. The method of claim 12 wherein the mammalian  
5 RNase H polypeptide is a cloned and expressed mammalian RNase H polypeptide.

19. A method of eliciting cleavage of a selected cellular RNA target comprising:

(a) providing an antisense oligonucleotide targeted  
10 to a selected RNA target to be cleaved;

(b) allowing said oligonucleotide and said RNA target to hybridize to form an oligonucleotide-RNA duplex; and

(c) contacting said oligonucleotide-RNA duplex with  
15 an RNase HI polypeptide having SEQ ID NO: 6, 7, 8, 9 or 11, under conditions in which cleavage of the RNA strand of the oligonucleotide-RNA duplex occurs,

whereby cleavage of the cellular RNA target is elicited.

20. The method of claim 19 wherein the RNase HI polypeptide is a cloned and expressed RNase HI polypeptide.

21. A method of eliciting cleavage of a selected cellular RNA target comprising:

(a) providing an antisense oligonucleotide targeted  
25 to a selected RNA target to be cleaved;

(b) allowing said oligonucleotide and said RNA target to hybridize to form an oligonucleotide-RNA duplex; and

(c) contacting said oligonucleotide-RNA duplex with  
30 an RNase HII polypeptide having SEQ ID NO: 1 or 10, under conditions in which cleavage of the RNA strand of the

oligonucleotide-RNA duplex occurs,  
whereby cleavage of the cellular RNA target is  
elicited.

22. The method of claim 21 wherein the RNase HII  
5 polypeptide is a cloned and expressed RNase HII  
polypeptide.

23. A method of screening oligonucleotides to  
identify an effective antisense oligonucleotide for  
inhibition of expression of a selected target protein  
10 comprising:  
(a) contacting a mammalian RNase H polypeptide with  
an RNA encoding the selected target protein and an  
oligonucleotide complementary to at least a portion of the  
RNA under conditions in which an oligonucleotide-RNA duplex  
15 is formed;  
(b) detecting cleavage of the RNA of the  
oligonucleotide-RNA duplex wherein cleavage is indicative  
of antisense efficacy; and  
(c) determining the site on the RNA at which  
20 cleavage occurs, whereby said site is identified as a RNase  
H-sensitive site.

24. The method of claim 23 further comprising  
identifying an effective antisense oligonucleotide which  
hybridizes to said RNase H-sensitive site.

25. The method of claim 23 wherein the  
oligonucleotide is one of a mixture or library of  
oligonucleotides.

26. The method of claim 23 wherein the mammalian  
RNase H polypeptide is a human RNase H polypeptide.

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27. The method of claim 23 wherein the mammalian RNase H polypeptide is an RNase HI polypeptide.

28. The method of claim 23 wherein the mammalian RNase H polypeptide is an RNase HII polypeptide.

5           29. The method of claim 23 which is performed in cells or tissues.

30. The method of claim 23 which is performed in an animal.

10           31. The method of claim 23 which is performed in a cell-free system.

32. A method of screening oligonucleotides to identify an effective antisense oligonucleotide for inhibition of expression of a selected target protein comprising:

15           (a) contacting an enriched amount of a mammalian RNase H polypeptide with an RNA encoding the selected target protein and an oligonucleotide complementary to at least a portion of the RNA under conditions in which an oligonucleotide-RNA duplex is formed; and

20           (b) detecting cleavage of the RNA of the oligonucleotide-RNA duplex wherein cleavage is indicative of antisense efficacy.

33. The method of claim 32 wherein the mammalian RNase H polypeptide is overexpressed or exogenously added.

34. The method of claim 32 wherein the mammalian RNase H polypeptide is an isolated, purified RNase H polypeptide.

35. The method of claim 32 wherein the mammalian  
5 RNase H polypeptide is a cloned and expressed RNase H polypeptide.

36. A method of prognosticating efficacy of antisense therapy of a selected disease comprising measuring the level or activity of a human RNase H  
10 polypeptide in a target cell of the antisense therapy.

37. The method of claim 36 wherein the human RNase H polypeptide is a human RNase HI polypeptide.

38. The method of claim 36 wherein the human RNase H polypeptide is a human RNase HII polypeptide.

15 39. A method of identifying agents which increase or decrease activity or levels of a mammalian RNase H polypeptide in a host cell comprising:

(a) contacting a cell expressing a mammalian RNase H polypeptide with an agent suspected or increasing or  
20 decreasing activity or levels of the mammalian RNase H polypeptide; and

(b) measuring the activity or levels of the mammalian RNase H polypeptide in the presence and absence of the agent so that an increase or decrease in the  
25 activity or levels of the mammalian RNase H polypeptide can be determined.

40. The method of claim 39 wherein the mammalian RNase H polypeptide is a human RNase H polypeptide.

41. The method of claim 39 wherein the mammalian RNase H polypeptide is an RNase HI polypeptide.

42. The method of claim 39 wherein the mammalian RNase H polypeptide is an RNase HII polypeptide.

5           43. An isolated human RNase HII polypeptide comprising SEQ ID NO: 1 or mutant form or active fragment thereof.

10           44. An isolated human RNase HII polypeptide encoded by the nucleotide sequence contained within ATCC Deposit No. PTA-2897 or mutant form or active fragment thereof.

45. A composition comprising a human RNase HII polypeptide and a pharmaceutically acceptable carrier.

15           46. An isolated polynucleotide encoding the human RNase HII polypeptide encoded by the nucleic acid sequence of the cDNA contained within ATCC Deposit No. PTA-2897, or mutant form or active fragment thereof.

47. The isolated polynucleotide of claim 46 which comprises SEQ ID NO: 12.

20           48. A vector comprising a nucleic acid encoding the human RNase H polypeptide encoded by the nucleic acid sequence of the cDNA contained within ATCC Deposit No. PTA-2897.

49. A host cell comprising the vector of claim 48.



50. A composition comprising a vector comprising a nucleic acid encoding a human RNase HII polypeptide and a pharmaceutically acceptable carrier.

51. An antibody targeted to the human RNase HII  
5 polypeptide of claim 44.

52. A nucleic acid probe capable of hybridizing to a portion of a nucleic acid encoding the polypeptide of claim 44.

53. A compound 8 to 50 nucleobases in length  
10 targeted to a nucleic acid molecule encoding a human RNase HII polypeptide, wherein said compound specifically hybridizes with and inhibits the expression of a human RNase HII polypeptide.

54. The compound of claim 53 which is an antisense  
15 oligonucleotide.

55. The compound of claim 54 wherein the antisense oligonucleotide has a sequence comprising at least an 8-nucleobase portion of SEQ ID NO: 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34 or 36.

56. The compound of claim 54 wherein the antisense  
20 oligonucleotide comprises at least one modified internucleoside linkage.

57. The compound of claim 56 wherein the modified internucleoside linkage is a phosphorothioate linkage.

58. The compound of claim 54 wherein the antisense  
25 oligonucleotide comprises at least one modified sugar moiety.

59. The compound of claim 58 wherein the modified sugar moiety is a 2'-O-methoxyethyl sugar moiety.

60. The compound of claim 54 wherein the antisense oligonucleotide comprises at least one modified nucleobase.

5 61. The compound of claim 60 wherein the modified nucleobase is a 5-methylcytosine.

62. The compound of claim 54 wherein the antisense oligonucleotide is a chimeric oligonucleotide.

63. A composition comprising the compound of claim  
10 53 and a pharmaceutically acceptable carrier or diluent.

64. The composition of claim 63 further comprising a colloidal dispersion system.

65. A method of inhibiting the expression of a human RNase HII polypeptide in cells or tissues comprising  
15 contacting said cells or tissues with the compound of claim 53 so that expression of the human RNase HII polypeptide is inhibited.

66. A method of treating an animal having a disease or condition associated with a human RNase HII  
20 polypeptide comprising administering to said animal a therapeutically or prophylactically effective amount of the compound of claim 53 so that expression of the human RNase HII polypeptide is inhibited.